Clinica Chimica Acta 481 (2018) 75–82

Contents lists available at ScienceDirect

Clinica Chimica Acta

journal homepage: www.elsevier.com/locate/cca

Monitoring opioid and benzodiazepine use and abuse: Is oral fluid or urine the preferred specimen type?☆

Athena K. Petrides[a,b,*], Stacy E.F. Melanson[a,b], Michalis Kantartjis[a,c], Rachel D. Le[d], Christiana A. Demetriou[e,f], James G. Flood[b,g]

[a] Department of Pathology, Brigham and Women's Hospital, Boston, MA, United States
[b] Harvard Medical School, Boston, MA, United States
[c] Department of Medicine, Brigham and Women's Hospital, Boston, MA, United States
[d] University of Massachusetts Medical School, Worcester, MA, United States
[e] The Cyprus Institute of Neurology and Genetics, Nicosia, Cyprus
[f] The Cyprus School of Molecular Medicine, Nicosia, Cyprus
[g] Department of Pathology, Massachusetts General Hospital, Boston, MA, United States

ARTICLE INFO

Keywords:
Oral fluid drug testing
Urine drug testing
Opioid
Benzodiazepine
Liquid chromatography-tandem mass spectrometry
Pain management

ABSTRACT

Background: Oral fluid (OF) has become an increasingly popular matrix to assess compliance in pain management and addiction settings as it reduces the likelihood of adulteration. However, drug concentrations and windows of detection are not as well studied in OF as in urine (UR). We compared the clinical utility and analytical performance of OF and UR as matrices for detecting common benzodiazepines and opioids.

Methods: OF and UR concentrations of 5 benzodiazepines and 7 opioids were measured by liquid chromatography-tandem mass spectrometry (LC-MS/MS) in 263 paired OF and UR specimens. UR creatinine was measured and prescription medications were reviewed.

Results: The benzodiazepines 7-aminoclonazepam, lorazepam, and oxazepam exhibited statistically higher detection rates in UR. For opioids, 6-AM was statistically more likely to be detected in OF, while hydromorphone and oxymorphone were statistically more likely to be detected in UR. Chemical properties including glucuronidation explain preferential detection in each matrix, not UR creatinine nor prescription status.

Conclusion: We found that OF is the preferred matrix for 6-AM, while UR is preferred for 7-aminoclonazepam, lorazepam, oxazepam, hydromorphone, and oxymorphone. However, OF should be considered if the risk of adulteration is high and use and/or misuse of benzodiazepines, hydromorphone, and oxymorphone is low.

1. Introduction

Between 2000 and 2015, half a million deaths were due to drug overdoses, and for the first time, in 2015, drug overdoses were the leading cause of accidental death in the United States, highlighting prescription drug misuse and addiction as a national issue in recent years [1,2]. Prescription opioids and benzodiazepines constituted nearly all (70% and 30%, respectively) of prescription overdose deaths in 2013, with deaths commonly involving both substances [1,3–5].As a consequence, substantial efforts have been made to prevent and treat substance abuse, including opioid-agonist medication-assisted treatment (OA-MAT) [1,2].

Routine and random drug testing as an adjunct to OA-MAT has provided an objective measure of compliance and treatment efficacy in both the pain management and addiction settings [6–8]. Historically, urine (UR) specimens have been used for drug monitoring. UR collection is non-invasive and drugs are present at higher concentrations for longer periods of time compared to serum [6–9]. Numerous studies have demonstrated the effectiveness of UR drug testing for monitoring compliance [6–8]. However, UR can be easily adulterated, particularly if collections are not observed, and patients with shy bladder or anuria may not be able to provide a specimen [10].

For this reason, the utility of oral fluid (OF) has been explored as a tool to assess compliance. OF collection significantly reduces the

Abbreviations: opioid-agonist medication-assisted treatment, (OA-MAT); oral fluid, (OF); urine, (UR); Massachusetts General Hospital, (MGH); liquid chromatography-tandem mass spectrometry, (LC-MS/MS); 6-acetylmorphine, (6-AM); mass spectrometer, (MS); Brigham and Women's Hospital, (BWH); heated electrospray ionization, (HESI); Substance Abuse and Mental Health Services Administration, (SAMHSA)

☆ This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

* Corresponding author at: Brigham and Women's Hospital, 75 Francis Street, Amory 2, Boston, MA 02115, United States.
E-mail address: apetrides@bwh.harvard.edu (A.K. Petrides).

https://doi.org/10.1016/j.cca.2018.02.034
Received 22 December 2017; Received in revised form 7 February 2018; Accepted 26 February 2018
Available online 27 February 2018
0009-8981/

likelihood of adulteration. In recent years, more studies have investigated OF in monitoring drugs of abuse with promising results. Nordal et al. reported that benzodiazepines can be measured qualitatively in OF and suggested OF may be an alternative for detection of clonazepam, diazepam, and alprazolam [11]. Likewise, Conermann et al. concluded that OF may be used for monitoring compliance of opioids and benzodiazepines after analyzing 132 paired specimens [12].

OF has some analytical challenges including low sample volumes and difficult specimen collections in patients with conditions such as dry mouth. Furthermore, collections are typically performed by clinic staff and can be time-consuming as they involve the patient rinsing their mouth and waiting 10–15 min prior to collection. Additionally, OF specimens may become contaminated with mouthwash, toothpaste, foods, and beverages [13,14]. Buccal contamination of OF by placing the drug sublingually immediately prior to collection may also pose a challenge for drug compliance assessment. Although OF has been studied less extensively, current literature demonstrates that OF exhibits lower drug and metabolite concentrations and narrower windows of detection compared to UR for the majority of drugs [9].

There are a limited number of studies directly comparing the performance of paired OF and UR specimens for both opioids and benzodiazepines. Additionally, to our knowledge, there are no published studies reporting the ratio of OF to UR concentrations for opioids and benzodiazepines in matched OF and UR specimens. In this study, we examined the clinical and analytical performance of each matrix and make suggestions on the utility of each matrix.

2. Methods

2.1. Specimen acquisition

A total of 263 paired OF and UR specimens were collected consecutively from 140 unique patients at the Massachusetts General Hospital (MGH) addiction-psychiatry clinics during routine visits and processed at the MGH Clinical Chemistry Laboratory (Boston, MA). For OF collection, the Orasure Intercept Sample Collection Device (Orasure Technologies, Bethlehem, PA) was utilized according to the manufacturer's collection instructions. OF and UR pairs were received by the laboratory within 2–10 h of collection. OF was refrigerated overnight and tested the next day. UR was frozen within 8 h of receipt and tested in batches at a later date. The Partners Human Research Committee approved this study.

2.2. Oral fluid drug analysis

As published previously, OF was analyzed at MGH using a laboratory-developed liquid chromatography-tandem mass spectrometry (LC-MS/MS) method [15,16]. The following benzodiazepines and opioids were detected: 7-aminoclonazepam, alprazolam, lorazepam, nordiazepam, oxazepam, 6-acetylmorphine (6-AM), codeine, hydrocodone, hydromorphone, morphine, oxycodone, and oxymorphone. Briefly, OF specimens were mixed with Internal Standard Solution containing deuterated analogues of each analyte. The mixture was injected onto a TLX2 chromatograph (Thermo Scientific, Waltham, MA) where the analytes and internal standards were first isolated on a Cyclone-P turbulent-flow extraction column (Thermo-Fisher, Franklin, MA) and then transferred to an Ascentis Phenyl analytical column (Supelco, Bellefonte, PA). The analytes were then separated using a gradient elution program and detected using a Thermo Quantum Ultra triple quadrupole MS (Thermo Scientific, Waltham, MA) equipped with a heated electrospray interface (HESI-II) operated in the positive ion mode. The limits of detection (LOD) for each drug or metabolite in OF are described in Table 1.

Table 1
Limit of detection (LOD) for urine (UR) versus oral fluid (OF) liquid chromatography-tandem mass spectrometry (LC-MS/MS) Testing.

Class	Drug or metabolite	LOD (ng/mL) for LC-MS/MS	
		UR	OF
Benzodiazepines	7-Aminoclonazepam	50	1
	Alpha-OH-Alprazolam	50	N/A
	Alprazolam	N/A	2
	Lorazepam	50	2
	Nordiazepam	50	2
	Oxazepam	50	2
Opioids	6-Acetylmorphine	5	2
	Codeine	50	2
	Hydrocodone	50	2
	Hydromorphone	50	4
	Morphine	50	2
	Oxycodone	50	1
	Oxymorphone	50	2

2.3. Urine drug analysis

UR was analyzed at the Brigham and Women's Hospital (BWH) Clinical Chemistry Laboratory (Boston, MA) using a laboratory-developed LC-MS/MS method. Samples were prepared by adding an internal deuterated standard to the following drugs or metabolites: 7-aminoclonazepam, alpha-hydroxy-alprazolam, lorazepam, nordiazepam, oxazepam, 6-AM, codeine, hydrocodone, hydromorphone, morphine, oxycodone, and oxymorphone. Samples were diluted and subjected to a hydrolysis step to remove glucuronide and sulfate groups. Chromatographic separation was achieved on a ACQUITY UPLC I-Class (Waters, Milford, MA) using a Kinetex C18 analytical column (Phenomenex Inc., Torrance, CA) and mass spectrometric analysis was performed on a tandem triple quadrupole Xevo TQS (Waters, Milford, MA) preceded by HESI. 6-AM was analyzed with the same equipment, but was not subjected to a hydrolysis step. The limits of detection (LOD) for each drug or metabolite in UR are also described in Table 1.

2.4. Calculation of oral fluid urine ratios

UR creatinine was measured using the rate-blanked Jaffe reaction with Roche Diagnostics reagents on a Roche Cobas e501 (Roche Diagnostics, Indianapolis IN). Quantitative UR drug measurements for 7-aminoclonazepam, alprazolam/alpha-hydroxy-alprazolam, nordiazepam, 6-AM, codeine, morphine, oxycodone, and oxymorphone were corrected for creatinine levels using the following formula: [Drug in ng/mL]/[UR Creatinine in mg/dL] × 100. Corrected drug concentrations were used to calculate oral fluid:urine ratios (OF:UR) using the following formula: [OF Drug in ng/mL]/[UR Drug in ng/mL]. Ranges and medians were calculated.

2.5. Medication review

The electronic health records of all patients in the study were reviewed for any active prescriptions/medications at the time of specimen collection. In this study, a prescription was considered active if the start date was before specimen collection and the end date was after or within 5 days of specimen collection.

2.6. Statistical analysis

The non-parametric McNemar's symmetry test was used to assess agreement between paired OF and UR specimens. A p-value < 0.05 was considered to be statistically significant.

To assess for dilution of UR specimens, creatinine concentrations of UR specimens were calculated. The UR creatinine distribution in